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format. Such a two-compartment reactor may be operated in parallel for multiple drug screening or kinetic analysis.

EXAMPLE

Cell-to-Cell Interaction

Cell-to-cell in vivo interaction was seldom studied directly due to the lack of control in cell manipulation. The present invention provides a powerful tool in this area. As illustrated in FIG. **26**, cells may be temporally and spatially controlled to favor the binding between the cells. Alternatively, cells may be directed to preassembled cell arrays for cell binding analysis. It may also be possible to study the interaction between cells such as T cells, B cells and polymorphonulear leukocytes, in the activation of immune responses.

EXAMPLE

Chemotaxis Analysis

Cells can be positioned in a controlled configuration to test their chemotaxis properties. This model is illustrated in FIG. **27**. For example, *D. discoideum* cells may be tested under cAMP or specific polypeptide gradient. (Parent & Devreotes, "A cell's sense of direction", Science 284, 765–770 (1999); the contents of which are incorporated herein by reference). Response of the receptors, G proteins, actin or actin-binding proteins may be detected through fluorescence signal generated by their correspondent fusion protein with GFP (Green Fluorescent Protein). Movement of the cells may also be recorded. More importantly, the force of the movement may be measured by comparing to the energy used in the light control of the cells.

The present invention may be used for cell identification and detection, for example, in immunophenotyping and affinity cell detection. In this application, affinity probes are coupled to beads on a solid surface. The beads are chemically and spatially coded. The cells are transported to the desired position using light control. Cells with the surface markers will bind at the correspondent positions. The advantages of this approach include a simpler and less expensive system, a higher degree of throughput for parallel immunophenotyping, the manipulation of a large number of cells, thus the detection of rare events, and better integration between the reaction and the detection steps, thus allowing the entire process to be performed on-chip.

The present invention may also be used in cell sorting, fractionation and isolation. In this case, cells can be separated according to their intrinsic properties, such as their dielectrophoretic mobility, viability, size, etc. Cells can also be sorted by their association with antibody coupled beads. The advantages of this approach include multiple cell sorting, simpler substrate fabrication, and flexible and programmable control.

The present invention may also be used for screening, for example, cell mediated drug and ligand screening, or cell secretion mediated screening. In this approach, cells may be transported and assembled a the desired position. Chemical compounds or ligands are released at the corresponding position. The response of the cells may then be observed through their reporter genes. The advantages of this approach include higher throughput, a simpler system without the complication of automated pipetting machines, and the possible combination with microscopy for simpler and more direct assays.

The present invention may also be used for cell to cell interaction, for example, in suspension or on surface interaction. In this approach, cells may link with each other through their surface interaction. Movement of the cells may then be guided by light. The advantages of this approach include the ability to study the interaction between cells.

While the invention has been particularly shown and described with reference to a preferred embodiment thereof, it will be understood by those skilled in the art that various changes in form and details may be made therein without departing from the spirit and scope of the invention.

What is claimed is:

1. A method of forming an assembly of cells in a designated area on a light-sensitive electrode, comprising:

providing a plurality of cells suspended at an interface between said light-sensitive electrode electrolyte solution;

generating an electric field at the interface; and

illuminating the interface with a predetermined light pattern to form a planar assembly of substantially one layer of cells in a designated area on the light-sensitive electrode, wherein the designated area is defined by the pattern of illumination.

2. The method of claim **1**, further comprising an additional electrode, the additional electrode and the light-sensitive electrode being substantially planar and aligned to one another and separated by a gap, wherein the additional electrode comprises an optically transparent electrode and wherein the gap contains the plurality of cells.

3. The method of claim **1**, wherein the electrode has a surface or an interior, the surface or interior having been modified to produce spatial modulations in properties of the electrode, said properties affecting the local distribution of the electric field at said interface, wherein the surface or interior have been modified by spatially modulated oxide growth, surface chemical patterning or surface profiling.

4. The method of claim **1**, further comprising the step of spatially or temporally varying the light pattern to cause the alteration of the assembly, said alteration being selected from the group consisting of disassembly, disassembly followed by reassembly, repositioning of the assembly, reconfiguration of the assembly, and segmentation of the assembly.

moving

5. The method of claim **1**, further comprising the step of varying the frequency or the voltage of the electric field to cause the alteration of the assembly, said alteration being selected from the group consisting of disassembly, disassembly followed by reassembly, repositioning of the assembly, reconfiguration of the assembly, and segmentation of the assembly.

6. The method of claim **1**, in which the plurality of cells comprises more than one type of cells, the method further comprising the step of fractionating one type of cells from another to induce a displacement of the plurality of cells within the assembly, fractionation arising as a result of differences in mobility of said types of cells.

7. The method of claim **1**, further comprising the step of maintaining the cells in the assembly, wherein the maintenance step comprises maintaining the electric field and the predetermined light pattern, or immobilizing the cells by chemical or physical means.

8. The method of claim **7**, in which the cellular assembly is immobilized on the electrode by chemically linking the cells or confining the cells.

9. An assembly of cells formed in a designated area on a light-sensitive electrode, comprising

the light-sensitive electrode; and

a planar assembly of cells comprising substantially one layer of cells in said designated area on the light-sensitive electrode, wherein the assembly is formed according to the method of claim **1**.

10. A method of conducting a bioassay involving an assembly of cells, comprising:

providing a planar assembly of cells comprising substantially one layer of cells in a designated area on the light-sensitive electrode and wherein the assembly of the cells is formed according to the method of claim 1;

contacting the cells with an analyte; and

detecting the binding of the analyte to the cells.

11. The method of claim 10, in which the cells in the assembly of the cells are immobilized prior to contacting the cells with the analyte.

12. The method of claim 11, in which the cells are immobilized by chemically linking the cells or confining the cells.

13. The method of claim 10, in which the analyte is directed to a specific cellular marker, and the bioassay is for determining the presence of the cellular marker on the surface of the cells.

14. The method of claim 13, in which the bioassay is directed to cell typing, with the presence of the marker on the cell surface indicating the cell type.

15. The method of claim 13, wherein the analyte is attached to a label and the detection of the binding of the analyte to the cells is carried out by detecting the presence of the label.

16. The method of claim 13, further comprising the step of removing the analyte that is not bound to the cell before the detection step is carried out.

17. The method of claim 13, wherein the analyte is attached to a label, said label comprising a flourescent tag.

18. The method of claim 13, wherein the analyte is attached to a label, said label comprising a bead which is distinguishable by chemical or physical characteristics.

19. The method of claim 13, in which more than one analyte is tested simultaneously for binding with the cells, with each analyte being attached to an encoded bead that is distinguishable by chemical or physical characteristics, and the detecting step comprising decoding the beads bound to the cells to determine the respective identities of analytes bound to the cells.

20. The method of claim 13, wherein the analyte is a ligand directed to a specific cellular receptor and the bioassay is for determining the presence of the receptor on the surface of the cells.

21. The method of claim 13, wherein the analyte is an antibody directed to a specific cellular antigen, and the bioassay is for determining the presence of the antigen on the surface of the cells.

22. The method of claim 13, in which the presence of more than one antigen is determined using more than one antibody, wherein each antibody is attached to a fluorophore tag that is chemically distinguishable, wherein the detection step comprises multicolor imaging of the cells.

23. A method of assaying the binding of cells with a ligand or an antibody, said method involving a planar assembly of cells and comprising:

providing an assembly of cells prepared according to claim 1, wherein the assembly further comprises a plurality of beads randomly mixed with the cells in the designated area on the light-sensitive electrode, said beads having a ligand or antibody attached to their surfaces, wherein said beads comprise different types of beads distinguishable by the ligand or antibody attached thereto and further distinguishable by a unique chemical or physical characteristic that identifies said bead type;

allowing the binding interaction to occur between the cells and the ligand or antibody;

disassembling the mixed assembly of the cells and the beads; and

detecting the binding interaction by analyzing the formation of clusters composed of the cells and the beads, the binding indicating the presence or absence of a cellular receptor or an antigen specific for the ligand or the antibody.

24. A method of determining a cellular response to an analyte, the method involving an assembly of cells and comprising: providing a planar assembly of cells formed according to claim 1; contacting the cells with an analyte; and detecting a cellular response to the analyte.

25. The method of claim 24, wherein the analyte comprises a drug molecule or a ligand.

26. The method of claim 24, in which the cellular response being detected is an expression of a particular gene, said expression being determined by detecting for the presence of an intracellular reporter gene product.

27. The method of claim 26, in which the expression of the intracellular reporter gene yields intracellular fluorescence.

28. The method of claim 24, in which the cellular response being detected is selected from the group consisting of: morphological change of the cells, change in the cell membrane permeability, and a change in the chemiotaxis response by monitoring the movement of cells.

29. A method of detecting a cellular response to an analyte, the method involving an assembly of cells and comprising: providing a planar assembly of cells prepared according to the method of claim 1; contacting the cells with an analyte; and detecting a cellular response to the analyte.

30. The method of claim 29, wherein one or more analytes are being tested for its ability to induce the cells to secrete one or more biologically active substances, and the detection step comprises detecting the presence of the biologically active substances.

31. The method of claim 30, wherein the biologically active substance comprises cytokine.

32. A method of determining a cellular response to an analyte, the method comprising:

providing a first electrode positioned in a first plane and a second electrode positioned in a second plane different from the first plane, the second electrode comprising a light-sensitive electrode and the first and the second electrode each comprising a planar electrode and said electrodes being in a substantially parallel alignment and separated by a gap, said gap containing an electrolyte solution in which a plurality of cells is suspended;

providing a planar array of beads on the first electrode, said beads having a ligand attached to their surfaces in a releasable manner, wherein said beads comprise different types of beads distinguishable by the ligand attached thereto and further distinguishable by a unique chemical or physical characteristic that identifies said bead type

illuminating an interface between the light-sensitive electrode and an electrolyte solution with a predetermined light pattern and generating an electric field at said interface to form a planar assembly of the cells on the second electrode, with the gap area separating the beads from the assembled cells; and

releasing the ligand and monitoring a cellular response, the proximity of the cells to the bead array permitting determination of the identity of the ligand inducing the cellular response.

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point at zero velocity, the shed coil will be fixed in position This method has several advantages over the prior art approaches. First, DNA molecules in their coiled state are subjected to light control to form arrays of desired shape in any position on the surface. This is possible because large DNA from cosmids or YACs forms coils with a radius in the range of one micron, and thus acts in a manner analogous to colloidal beads. A set of DNA molecules may thus be steered into a desired initial arrangement. Second, UV-patterning ensures that the elongational force created by the electrokinetic flow is directed in a predetermined direction. The presence of metal electrodes in contact with the sample, a disadvantage of the dielectrophoretic prior art method, is avoided by eliminating this source of contamination that is difficult to control especially in the presence of an electric field. On patterned Si/SiOx electrodes, flow velocities in the range of several microns/second have been generated, as required for the elongation of single DNA molecules in flow. Thus, gradients in the flow field determines both the fractional elongation and the orientation of the emerging linear configuration. Third, the present invention facilitates direct, real-time control of the velocity of the electric field-induced flow, and this in turn conveys explicit control over the fractional elongation.

While the invention has been particularly shown and described with reference to a preferred embodiment thereof, it will be understood by those skilled in the art that various changes in form and details may be made therein without departing from the spirit and scope of the invention.

What is claimed is:

1. A method for moving particles suspended at an interface between an electrode and electrolyte solution comprising the following steps:

providing a first electrode positioned in a first plane, and a second electrode positioned in a second plane different from the first plane, an electrolyte solution located therebetween and a plurality of particles suspended in the electrolyte solution, wherein the second electrode comprises a planar electrode having a surface and an interior, the surface or interior having been modified to produce spatial modulations in electrochemical properties of the second electrode; and

generating an electric field at an interface between the electrolyte solution and the second electrode to effect movement of the particles, wherein the movement of the particles at the interface is in accordance with the electric field in combination with the spatial modulations in the properties of the second electrode, said properties affecting the local distribution of the electric field at the interface.

2. The method of claim **1**, wherein said patterning step is used to create one or more areas of low impedance on said electrode.

3. The method of claim **2**, wherein at least one of the frequency or magnitude of the applied voltage is adjusted so as to cause said particles to move preferentially into or out of one or more of said areas of low impedance.

4. The method of claim **1**, wherein said electrode includes a silicon electrode which is coated with a dielectric layer.

5. The method of claim **1**, wherein the spatial modulations of the properties of the second electrode are produced by patterning the surface or the interior of the second electrode by spatially modulated oxide growth, surface chemical patterning or surface profiling.

6. The method of claim **1**, wherein the first electrode and the second electrode each comprises a planar electrode, said first and second electrodes being parallel to another and separated by a gap, with the electrolyte solution containing the particles being located in the gap.

7. The method of claim **1**, wherein the property of the second electrode being modulated comprises impedance, one or more areas of the surface or the interior of the second electrode being modified to exhibit low impedance, and wherein the particles move to the areas of low impedance.

8. The method of claim **1**, wherein the electric field is generated by applying an AC voltage between the first and the second electrode.

9. The method of claim **1**, wherein the second electrode comprises a light-sensitive electrode, the method further comprising the step of illuminating the second electrode with a predetermined light pattern.

10. The method of claim **1**, wherein the movement of the particles is in a direction substantially parallel to the second electrode.

11. A method for controlling the movement of an electrolyte solution comprising the following steps:

providing a first electrode positioned in a first plane, and a second electrode positioned in a second plane different from the first plane, and an electrolyte solution located therebetween, wherein the second electrode comprises a planar electrode having a surface and an interior, the surface or interior having been modified to produce spatial modulations in electrochemical properties of the second electrode; and

generating an electric field at an interface between the electrolyte solution and the second electrode to create fluid flow of the electrolyte solution having a velocity, said velocity having a magnitude determined by said electric field and a direction determined by said spatial modulations in the properties of the second electrode, said properties affecting the local distribution of the electric field at the interface.

12. The method of claim **11**, wherein the spatial modulations of the properties of the second electrode are produced by patterning the surface or the interior of the second electrode by spatially modulated oxide growth, surface chemical patterning or surface profiling.

13. The method of claim **11**, wherein the property of the second electrode being modulated comprises impedance, one or more areas of the surface or the interior of the second electrode being modified to exhibit low impedance, and wherein the electrolyte solution moves to the areas of low impedance.

14. The method of claim **11**, wherein the first electrode and the second electrode each comprises a planar electrode, said first and second electrodes being parallel to another and separated by a gap, with the electrolyte solution being located in the gap.

15. The method of claim **11**, wherein the electric field is generated by applying an AC voltage between the first and the second electrode.

16. The method of claim **11**, wherein the second electrode comprises a light-sensitive electrode, the method further comprising the step of illuminating the second electrode with a predetermined light pattern.

17. The method of claim **11**, wherein the second electrode comprises a silicon electrode.

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the entire repertoire of beads in the planar army or panel formed adjacent to the electrode surface for parallel readout. As with heterogeneous panels in general, the arrangement of beads within the array is either random (with respect to chemical identity), and the identity of beads scoring high in the binding assay must be determined subsequently, or it is spatially encoded by invoking the "Layout-Preserving Transfer" method of sample loading described herein.

The former valiant is readily implemented and accommodates array formation either prior to or subsequent to performing the binding assay. For example, binding may be performed in suspension before beads are assembled into the array. As with the aforementioned cDNA selection procedure, the method of the present invention also accommodates the use of beads as capture elements for end-functionalized target DNA, for example, via biotin-streptavidin complexation. In this later case, beads serve as a delivery vehicle to collect all probe-target complexes to the electrode surface where they are assembled into an array for ease of analysis. In particular, proximity CCD detection of beads on electrodes will benefit from the lensing action of the beads in the array. This version of the assay is preferably used if only a small number of positive scores are expected.

Hybridization to a pre-formed bead array can take advantage of a variant of the assay which preserves spatial encoding. An array of bead clusters is formed by the "Layout-Preserving Transfer" method previously described herein, and exposed to a mixture of cDNAs. The resulting spatial distribution of fluorescence intensity or radioactivity reflects the relative abundance of cDNAs in the mixture. This procedure relies on the detection of a characteristic fluorescence or other signal from the probe-target complex on the surface of a single bead. Given the fact that the array is readily held stationary by the methods of the present invention, image acquisition may be extended to attain robust signal-to-noise for detection of low level signals. For example, a signal generated by a bead of 10 micron diameter with at most 10^8 probe-target complexes on the surface of the bead may be detected. Bead lensing action also aids in detection.

As with the implementation of drug screening, the functional elements of the present invention may be combined to perform multiple preparative and analytical procedures on DNA.

EXAMPLE X

Alignment and Stretching of DNA in Electric Field-Induced Flow

The present invention can be used to position high-molecular weight DNA in its coiled configuration by invoking the fundamental operations as they apply to other colloidal particles. However, in addition, the electrokinetic flow induced by an electric field at a patterned electrode surface may be employed to stretch out the DNA into a linear configuration in the direction of the flow.

Procedures have been recently introduced which rely on optical imaging to construct a map of cleavage sites for restriction enzymes along the contour of an elongated DNA molecule. This is generally known as a "restriction map". These procedures, which facilitate the study of the interaction of these and other proteins with DNA and may also lead to the development of techniques of DNA sequencing, depend on the ability to stretch and align DNA on a planar substrate.

For individual DNA molecules, this has been previously achieved by subjecting the molecule to elongational forces such as those exerted by fluid flow, magnetic fields acting on DNA-anchored magnetic beads or capillary forces. For example, DNA "combs" have been produced by simply placing DNA molecules into an evaporating droplet of electrolyte. If provisions are made to promote the chemical attachment of one end of the molecule to the surface, the DNA chain is stretched out as the receding line of contact between the shrinking droplet and the surface passes over the tethered molecules. This leaves behind dry DNA molecules that are attached in random positions within the substrate area initially covered by the droplet, stretched out to varying degrees and generally aligned in a pattern of radial symmetry reflecting the droplet shape. Linear "brushes", composed of a set of DNA molecules chemically tethered by one end to a common line of anchoring points, have also been previously made by aligning and stretching DNA molecules by dielectrophoresis in AC electric fields applied between two metal electrodes previously evaporated onto the substrate.

The present invention invokes electrokinetic flow adjacent to an electrode patterned by UV-mediated regrowth of oxide to provide a novel approach to the placement of DNA molecules in a predetermined arrangement on a planar electrode surface, and to the stretching of the molecules from their native coil configuration into a stretched, linear configuration that is aligned in a predetermined direction. This process is shown in FIG. 11 and is accomplished by creating controlled gradients in the flow vicinity across the dimension of the DNA coil. The velocity gradient causes different portions of the coil to move at different velocities thereby stretching out the coil. By maintaining a stagnation point at zero velocity, the stretched coil will be fixed in position. This method has several advantages over the prior art approaches. First, DNA molecules in their coiled state are subjected to light control to form arrays of desired shape in any position on the surface. This is possible because large DNA from cosmids or YACs forms coils with a radius in the range of one micron, and thus acts in a manner analogous to colloidal beads. A set of DNA molecules may thus be steered into a desired initial arrangement. Second, UV-patterning ensures that the elongational force created by the electrokinetic flow is directed in a predetermined direction. The presence of metal electrodes in contact with the sample, a disadvantage of the dielectrophoretic prior art method, is avoided by eliminating this source of contamination that is difficult to control especially in the presence of an electric field. On patterned Si/SiOx electrodes, flow velocities in the range of several microns/second have been generated, as required for the elongation of single DNA molecules in flow. Thus, gradients in the flow field determines both the fractional elongation and the orientation of the emerging linear configuration. Third, the present invention facilitates direct, real-time control of the velocity of the electric field-induced flow, and this in turn conveys explicit control over the fractional elongation.

While the invention has been particularly shown and described with reference to a preferred embodiment thereof, it will be understood by those skilled in the art that various changes in form and details may be made therein without departing from the spirit and scope of the invention.

What is claimed is:

1. A method for controlling the movement of particles suspended at an interface between an electrode and an electrolyte solution, the method comprising the following steps:

generating an electric field at said interface between said electrode and said electrolyte solution; and

illuminating the surface of said electrode with a predetermined light pattern to control the movement of said

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particles in accordance with said predetermined light pattern and electrochemical properties of said electrode.

2. The method of claim 1, wherein said electric field is at least one of a constant and a time varying electric field.

3. The method of claim 1, further comprising a patterning step which is performed using at least one of UV-mediated oxide regrowth, surface chemical patterning and surface charge profiling.

4. The method of claim 3, wherein said patterning step is used to create a plurality of areas of low impedance on said electrode, and said illuminating step is used to selectively connect one or more of said areas of low impedance to cause said particles to move therebetween in accordance with said patterning and said predetermined light pattern.

5. The method of claim 1, wherein the illuminating step comprises the further step of:

illuminating a selected area of said electrode which in conjunction with a frequency of said electric field causes the particles to move into said selected area.

6. The method of claim 5, wherein the frequency of said electric field is adjusted in order to move said particles out of said selected area.

7. The method of claim 1, wherein the illuminating step comprises the further step of:

illuminating a selected area of said electrode surface with a high intensity light pattern so as to cause the particles to move out of said selected area.

8. The method of claim 1 further comprising a patterning step which creates at least two areas of low impedance on said electrode, and said illuminating step being used to selectively cause said particles to move from a first low impedance area to a second low impedance area.

9. An apparatus for implementing the differential lateral displacement of particles suspended at an interface between an electrode and an electrolyte solution, said apparatus comprising:

an electric field generator which generates an electric field at said interface;

an electrode;

an electrolyte solution having a substantially continuous flow which effects the displacement of said particles in a direction substantially parallel to said interface;

an illumination source which illuminates said electrode with an adjustable, predetermined light pattern; and

a plurality of particles located in said electrolyte solution, said particles being in said electrolyte flow and being displaced by said electric field in conjunction with said predetermined light pattern, said particles being displaced in accordance with variations in physical and chemical properties of said particles which determine the mobility of said particles.

10. The apparatus of claim 9, wherein said electrode is a light sensitive electrode.

11. The apparatus of claim 9, wherein said impedance profile is created by a predetermined illumination pattern.

12. The apparatus of claim 9, wherein:

said electrode patterning includes an area of low impedance bordered by an area of high impedance, said low impedance area including a narrow conduit in communication with a wide conduit, both said conduits being oriented parallel to the direction of said continuous flow of said electrolyte;

said wide conduit including a row of intermittently spaced areas of high impedance barriers traversing the width of said wide conduit;

a portion of said plurality of particles being optically distinguishable from the remaining particles;

a detector for visually inspecting said particles traversing the length of said narrow conduit in response to said continuous flow of electrolyte;

said illumination pattern being substantially in the shape of a rectangle having a longer dimension adjusted to be substantially equal to the width of said wide conduit, said rectangle having a smaller dimension which is adjusted to be substantially equivalent to the diameter of said particles, said pattern being located in front of said barriers, and said illumination pattern conforming to an intensity profile placing a maximal value of intensity in the center of said wide conduit and decreasing symmetrically to lower values of intensity at the two sides of said wide conduit; and

a delay activation circuit which activates said illumination profile in response to a signal derived from said visual inspection of said particles so as to cause an illuminated particle to be displaced from regions of maximum intensity to regions of lower intensity of said intensity profile and to be deflected into the intermittent spaces between said barriers.

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